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Microbial Pigments for Sustainable Textile Dyeing

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ABSTRACT

Synthetic dyes have been the traditional choice for the textile industry to impart color, leading to major environmental issues because of chemical pollution and the release of dye contaminated effluents into the environment. Microbiological pigments have become a subject of interest as alternative eco-friendly colourants due to the growing awareness on Sustainable Manufacturing. These pigments are produced by bacteria, fungi, yeasts and microalgae and have benefits of renewability, biodegradability and comparatively less environmental impact. In addition to the coloration, some microbial pigments have functional properties, such as antimicrobial, antioxidant activity and protection from ultraviolet (UV) radiation, which could be used in multifunctional applications in textiles. In the present review, the sources, classification, production, extraction and purification of microbial pigments have been discussed, as well as their applications in the textile dyeing and printing industry. It also emphasizes how they interact with natural and synthetic fibres, their functional characteristics and their environmental value. In addition, issues of production costs, pigment stability, colour fastness, scalability and regulatory requirements are discussed. The developments in the scientific field of biotechnology, nanotechnology and sustainable bioprocessing provide possibilities to make them more industrial and to facilitate sustainable textile colouration.

Keywords: Microbial pigments, Sustainable textile dyeing, Bio-based colourants, Textile coloration

INTRODUCTION

The textile sector is one of the biggest and most important industries in the world and plays a crucial role in economic development, job creation and international trade. Textiles colouring is of great importance in improving the aesthetic, value and acceptance of the textile products. Therefore, dyeing and printing processes are an essential step in textile wet processing which enable the textile fabrics to be made in different colours, shades and decoration effects in the field of apparel, home furnishing, medical textile and technical textile. Hence, dyeing and printing process is also regarded as an essential step in the textile wet processing which enable the textile fabrics to be made in various colours, shades and decoration effects which are required in the field of apparel, home furnishing, medical textile and technical textile. The textile industry has depended on synthetic dyes, owing to the availability of a large number of colours, high colour strength, ease of application and the relatively low production costs. In the textile industry, various synthetic dyes are widely used, including azo dyes, reactive dyes, vat dyes, disperse dyes and sulfur dyes, which are applied to provide long-lasting and even coloration of different fibre types (Kant, 2012).

While synthetic dyes may have commercial benefits, the amount of water, chemicals, salts and auxiliary agents used in textile dyeing operations has raised significant issues regarding the environment and health. A significant amount of synthetic dyes is not bound to fiber during the dyeing process, and is released during washing and rinsing into the wastewater. Therefore, the textile dyeing industries are known to be among the significant industrial water polluting industries globally (Yaseen and Scholz, 2019). The effluents generated by the textile industry have been found to have high content of synthetic dyes,

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recalcitrant organic compounds which are not degraded naturally, high chemical oxygen demand (COD), biological oxygen demand (BOD) and toxicity (Holkar *et al.,* 2016). Therefore, the discharge of untreated wastewater containing dyes to aquatic ecosystems can lead to disturbance of the ecological balance, decrease in dissolved oxygen, depression of photosynthesis and impact on aquatic organisms and human health (Berradi *et al.,* 2019).

With an increasing demand for environmental awareness and tougher environmental laws, researchers and the textile industry have been looking for sustainable alternatives to traditional synthetic dyes. Microbial pigments have emerged as an eco-friendly dyes sources in recent years, since they are biodegradable, renewable and can be synthesized by environmentally friendly biotechnology methods. Various natural pigments have been synthesized by microorganisms like bacteria, fungi, yeasts and microalgae that have different colours and functional properties (Ramesh *et al.,* 2019). Apart from coloration, these pigments could also have antimicrobial, antioxidant, ultraviolet (UV) protection, and bioactive properties which can impart multifunctional properties to the textile products (Venil *et al.,* 2020).

As sustainable methods of microbial pigment production are not dependent on seasonal fluctuations or the availability of agricultural land, microbial fermentation has made microbial pigments an interesting target for sustainable textile dyeing. The microbial pigments can be produced more quickly than plant dyes, can be reproducible and less polluting than plant dyes (Mapari *et al.,* 2010). The potential of microbial pigment production by fermentation processes and its commercial application in textile industries has been further enhanced by the advances in industrial biotechnology, metabolic engineering and fermentation technology (Singh *et al.,* 2021).

Yet, few obstacles exist for the industrial use of microbial pigments in textile dyeing applications, which have a tremendous potential. The following topics are still relevant for research: aspect of pigment yield, production cost, colour fastness, pigment extraction efficiency and process standardization on a large scale. Hence, it is necessary to understand the potential, constraints and future of microbial pigments to design sustainable textile colouration system that will not be burdening the environment, yet will satisfy the performance standards for the industry.

Importance of Sustainable Textile Dyeing

The demand for sustainable textile dyeing processes has been growing significantly in recent decades due to the high level of pollution to the environment, high consumption of water and high consumption of chemicals in conventional dyeing processes. Textile industry consumes vast quantities of fibres and fabrics annually and dyeing processes consume massive quantities of dyes, salts, alkalis, acids, surfactants and auxiliary chemicals to obtain desirable colour properties and fastness characteristics (Shahid and Mohammad, 2013). Textile dyeing is one of the most problematic areas of textile production for its use of large amounts of water and the amount of wastewater it produces that contains hazardous pollutants.

Due to the growing demands for sustainable industrial production, eco-friendly technologies in textile processing that reduce environmental impact while retaining product quality and industrial efficiency have developed. The focus of sustainable textile dyeing is to minimize the use of water, chemicals, amount of toxic effluent released and to use biodegradable and renewable resources. It is important to use sustainable dyeing technologies especially as water scarcity, environmental degradation and pollution control are important global issues in both developed and developing countries.

Traditional dyes are a source of toxic aromatic chemicals, heavy metals and non-biodegradable dyes which can be present in the environment for a considerable time. Synthetic dye molecules have complicated aromatic structures that cannot be degraded by microorganisms in the aquatic environment after discharge into water bodies. These dyes have been known to be environmentally persistent, causing concern for environmental toxicity, mutagenicity and carcinogenicity. Furthermore, in the textile dyeing processes, many washing cycles are often needed and high temperatures are requested to colour the textiles, which leads to high energy consumption and greenhouse gas emissions (Niinimäki *et al.,* 2020).

The goal of sustainable textile dyeing is to build cleaner production systems, which aim to minimize the generation of pollution at the source, and not only by the application of end-of-pipe wastewater treatment technologies. The use of bio-available dyes, enzymatic processing, water-friendly dyeing systems, and environmentally friendly chemicals has become a significant focus of green textile dyeing strategies (Kate

et al., 2021). Microbial pigments have proven to be very promising alternatives among them since they can be synthesized in a fermentation process with environmentally friendly conditions, renewable raw materials, and relatively mild processing conditions.

Consumers' awareness of environmental-friendly textile products is another crucial factor in sustainable textile dyeing. Today's consumers are demanding eco-friendly products in sustainable fashion that are made with sustainable materials and green processing technology. Therefore, more and more attention is being paid to the sustainable dyeing techniques of textile industries, which should be in line with international environmental standards and the principles of circular economy (Niinimäki *et al*., 2020). Therefore, use of microbial pigments in textile dyes may not only help in protecting the environment, but can also be a way of increasing the competitiveness of the market and sustainable industrial development.

Environmental Concerns of Synthetic Dyes

Synthetic dyes are extensively used in textile industries due to their excellent colour brightness, wide range of shades, high colour fastness and cost-effectiveness. However, the environmental impacts with the production, application and disposal of these have become a major global concern. From the dyeing processes of textile industry, large amounts of coloured wastewater containing residual dyes, salts, surfactants, heavy metals and other toxic chemicals are discharged into the environment. It is estimated that about 10–15% of the synthetic dyes used in the textile dyeing processes are lost in wastewater streams during washing and rinsing operations (Holkar *et al*., 2016).

One of the most serious environmental problems associated with synthetic dye is water pollution. Discharge of effluents containing dyes into rivers and lakes reduces the transparency of water and penetration of sunlight into the aquatic ecosystems. The reduced light penetration affects the photosynthesis activity of aquatic plants and algae, disturbing the aquatic food chains and the stability of the ecosystem (Berradi *et al*., 2019). Synthetic dyes are designed to resist fading and degradation in environmental conditions and even very low concentrations of dyes can impart intense colour to water.

Additionally, a large number of synthetic dyes have complex aromatic molecular structures that make them resistant to microbial degradation. Consequently, they can remain in the aquatic environment long, and bioaccumulate in sediments and biological systems. Some azo dyes may be reductively cleaved under anaerobic conditions to yield aromatic amine producing some which are known to be cancer and mutating agents (Kant, 2012). Such exposures over extended periods can then be very harmful to aquatic life and human health.

The water from the textile dyeing process is also known to contain high concentrations of chemical oxygen demand (COD) and biological oxygen demand (BOD), which indicate high organic pollution. Untreated wastewater enters natural water bodies, and microorganisms use up the dissolved oxygen in taking up the organic pollutants, which leads to depletion of dissolved oxygen and harmful effects on aquatic life (Yaseen and Scholz, 2019). In extreme instances, when oxygen is lacking, fish can die and cause ecological imbalance.

Dyeing and manufacturing of synthetic dyes are not only the causes of aquatic pollution but also cause air pollution and soil contamination. Disposal of dye sludge obtained from the wastewater treatment plants can result in deposition of heavy metals and persistent organic pollutants in soils which can affect soil fertility and agricultural productivity. Furthermore, volatile organic compounds and toxic gases emitted during the manufacture of dyes and processing of textiles may also cause pollution of the atmosphere and occupational health hazards to textile workers (Lellis *et al*., 2019).

Given these environmental and health issues, governments and environmental agencies have introduced tougher limits on the discharge of textile wastewater and the use of chemicals in the industry throughout the world. Hence, there is a growing trend to replace the outdated and less environmentally friendly conventional textile technologies with safer and environmentally friendly technologies that minimize toxic emissions and promote cleaner production in the textile industry.

Need for Bio-based Colorants

The rising environmental concerns regarding synthetic dyes have generated a strong demand for eco-friendly and biodegradable alternatives in the textile colouration processes. Bio-based colourants are attractive alternatives since they are obtained from renewable biological sources and generally show lower toxicity and better biodegradability than synthetic dyes. These colourants can be obtained from plants, animals and microorganisms, but microbial pigments have been

recently much focused due to their great advantages in production and potential sustainability (Mapari *et al*., 2010).

It is closely related to the demand for bio-based colorants and the global trend towards sustainable industrial development and green manufacturing practices. The conventional production of synthetic dyes is energy-intensive and uses a high amount of petroleum resources, and complicated chemical synthesis routes produce hazardous by-products and environmental pollutants. Bio-based pigments, on the other hand, can be synthesized through microbial fermentation processes, using renewable organic raw materials, agricultural waste materials, and eco-friendly bioprocesses (Venil *et al*., 2020). This is in line with the concept of Circular Bioeconomy and less reliance on non-renewable fossil resources.

The rising consumer demand for eco-friendly textile products is another important factor contributing to the demand of bio-based colorants. The consumers are now increasingly aware of textile chemicals and their environmental and health effects and they are actively looking for sustainable fashion products made with natural and non-toxic chemicals. It is therefore imperative for textile brands and manufacturers to invest in green dyeing technologies to enhance the environmental performance and achieve sustainability targets (Niinimäki *et al*., 2020).

Microbial pigments are especially interesting as bio-based colourants since the microorganisms can produce a variety of pigments of different colours and properties. Carotenoids, melanins, prodigiosin, violacein, riboflavin, indigoidine are some of the pigments synthesized by bacteria, fungi, yeasts and microalgae. Several of these pigments also possess other functional characteristics like antimicrobial activity, antioxidant activity and ultraviolet protection, which are suitable for multifunctional applications in textile sector (Ravikumar *et al*., 2022).

Unlike plant derived natural dyes, microbial pigments can be produced rapidly under controlled fermentation conditions independent of seasonal variations, climatic conditions and availability of agricultural land. Cultivation of microbes also has advantages of shorter production cycles, easier genetic modification and better reproducibility of the process (Singh *et al*., 2021). The progress in biotechnology and metabolic engineering has further enhanced the feasibility of the improvement of pigment yield and optimisation of industrial-scale production processes.

However, despite these advantages, the commercial application of microbial pigments in textile dyeing is still hampered by several technical and economic challenges including pigment stability, efficiency of extraction, cost of production and colour fastness properties. However, research and technological advances still broaden the microbial pigments' horizon as sustainable alternatives for future textile colouration systems.

Microbial Pigments: Sources and Classification

Microbial pigments are natural colour compounds produced by microorganisms such as bacteria, fungi, yeast and microalgae during their metabolic activities. The pigments serve various physiological and ecological roles in microbial cells such as protection against ultraviolet radiation, oxidative stress, harsh environmental conditions and microbial competition. Microbial pigments have been of great interest to scientists and industries in recent years because of their potential applications in textiles, food, pharmaceuticals, cosmetics and biomedical industries (Mapari *et al*., 2010).

Microbial pigments are thus considered as sustainable and eco-friendly, in contrast to synthetic dyes which are mostly obtained from petrochemical sources, because of the possibility to produce them by fermentation processes with renewable substrates and relatively mild operating conditions. Therefore, the increasing importance of sustainable textile manufacturing has sped up the research on microbial pigments as alternative colourants for the textile dyeing applications (Venil *et al*., 2020).

Microorganisms are able to produce a wide variety of pigments of different chemical structures and colours. These pigments include carotenoids, melanins, flavins, quinones, prodigiosin, violacein and indigoidine etc. Microbial pigments may be different in colour and chemical composition for different microbial species, growth conditions, and availability of nutrients and environmental factors such as temperature and pH (Ramesh *et al*., 2019). Moreover, many microbial pigments have bioactivities such as antimicrobial, antioxidant, anticancer and ultraviolet protective activities, which are more suitable for functional textile applications.

Microbial pigments can be generally classified on the basis of the type of microorganism producing them or on the basis of their chemical composition and colour properties. Among the major microbial sources, bacteria, fungi, yeasts and microalgae are considered to be the

most important pigment-producing organisms for industrial applications.

Bacterial Pigments

Bacteria are one of the most studied microorganisms for pigment production due to their fast growth, simple nutritional requirements and ability to synthesise a wide range of coloured metabolites. The pigments of bacteria can be red, yellow, blue, violet, orange, and green, depending on the type of microbes and the structure of the pigments. Many bacterial pigments are secondary metabolites synthesised under specific environmental or stress conditions and may be involved in protection against oxidative damage, ultraviolet radiation or microbial competition (Venil and Lakshmanaperumalsamy, 2009).

Prodigiosin is one of the most important bacterial pigments that are produced mainly by Serratia marcescens in red colour. Prodigiosin has attracted much attention due to its antimicrobial, anticancer, antioxidant and immunosuppressive properties. The bright colour and biological functionality of the pigment make it a potential candidate for textile colouration (Darshan and Manonmani, 2015). Similar to that, violacein is a violet pigment produced by bacteria like *Chromobacterium violaceum* and *Janthinobacterium lividum.* Violacein shows antibacterial, antiviral, antifungal and anticancer activities, making it useful for multifunctional textile applications.

Several bacterial species, including Micrococcus, Rhodotorula and Paracoccus, also synthesise carotenoid pigments extensively. These pigments usually provide yellow, orange or red colours and show strong antioxidant activity, as they can neutralise free radicals and protect cells from oxidative stress (Mata-Gómez *et al*., 2014). Bacterial carotenoids are gaining interest as sustainable alternatives to synthetic colourants, due to their biodegradability and relatively low toxicity.

Pyocyanin, a blue-green pigment, is another important bacterial pigment produced by Pseudomonas aeruginosa. Pyocyanin has redox-active properties and is important for the competitive survival of the organism that produces it. Also, indigoidine, a blue pigment produced by some bacterial strains has been investigated as a natural alternative for blue textile dyes, as natural blue colourants are relatively rare in nature (Sajjad *et al*., 2020).

The production of bacterial pigment has several industrial advantages, such as obtaining of high growth rate, short time required for the fermentation process, ability to use inexpensive substrates, and the ease of genetic modification. The commercial interest of bacterial pigment production for textile dyes has been further enhanced through the development of advances in metabolic engineering and synthetic biology. The following table presents important bacterial pigments, their microbial sources, colour characteristics, and potential textile applications (Table 1).

Fungal Pigments

Another important group of microorganisms is fungi, which can produce a wide range of natural pigments of industrial importance. Pigment-producing fungi are filamentous fungi, moulds and some endophytic fungal species that produce coloured metabolites during their growth and sporulation processes. Fungal pigments have attracted much attention because fungi are able to produce large amounts of pigments on relatively inexpensive substrates under controlled fermentation conditions (Dufossé *et al*., 2014).

Fungal pigments, especially those from the Monascus species, are very important. Monascus purpureus produces red, orange and yellow pigments called Monascus pigments which have been traditionally used in Asian food products and are now being studied for textile colouration applications. These pigments exhibited good colour intensity and relative high stability

Table 1 : Important Bacterial Pigments and their Characteristics

Bacterial Species	Pigment Produced	Colour	Potential Applications	Reference
Serratia marcescens	Prodigiosin	Red	Textile dyeing, antimicrobial textiles	Darshan and Manonmani (2015)
Chromobacterium violaceum	Violacein	Violet	Functional textiles, biomedical applications	Venil *et al.* (2013)
Paracoccus carotinifaciens	Carotenoids	Orange/Red	Natural textile colorants, antioxidants	Mata-Gómez *et al.* (2014)
Pseudomonas aeruginosa	Pyocyanin	Blue-green	Bioactive textile coloration	El-Fouly *et al.* (2015)
Streptomyces spp.	Actinorhodin	Blue	Natural dyeing applications	Sajjad *et al.* (2020)

under various processing conditions (Mapari *et al*., 2010).

Many fungi such as Aspergillus, Penicillium, Fusarium and Talaromyces are known to produce a variety of coloured compounds such as anthraquinones, melanins, flavins and carotenoids. Fungal melanin pigments have strong UV protection and antioxidant properties that make them suitable for functional textile applications requiring UV protection (Gmoser *et al*., 2017).

Fungal pigments are promising candidates for sustainable textile dyeing, as the fungal cultivation can be based on agro-industrial wastes and low-cost substrates, which can reduce the production costs and support waste valorisation strategies. Also, fungal fermentation can result in higher concentrations of pigments than some bacterial systems. However, some fungal pigments may also contain mycotoxins or other undesired secondary metabolites, so that the appropriate strain selection and purification are necessary prior to their industrial use (Dufossé *et al*., 2014). The following table summarizes major fungal pigments and their relevance in sustainable textile coloration (Table 2).

Yeast and Microalgal Pigments

In recent years, yeasts and microalgae have been identified as natural resources that have the potential to produce natural pigments with high biological activity and applicability in the industry due to their sustainability. There are many benefits to these microorganisms including rapid growth, scalability and growth in a controlled environment.

Pigmented yeasts such as Rhodotorula, Phaffia rhodozyma and Yarrowia lipolytica are known to produce carotenoid pigments such as b-carotene, torulene and astaxanthin. These pigments are also useful for textile industry as they have high antioxidant activity and are capable of creating yellow to orange and red colors (Buzzini *et al*., 2017). In particular, astaxanthin has a high antioxidant capacity and commercial value in the nutraceutical and cosmetic industry.

In addition, microalgae are important producers of natural pigments such as chlorophylls, carotenoids and phycobiliproteins. Species such as Spirulina, Chlorella, *Dunaliella salina*, and *Haematococcus pluvialis* are extensively investigated for pigment production due to their ability to accumulate high levels of bioactive pigments when subjected to stress conditions (Morales-Oyervides *et al*., 2020). Since the stable natural blue colourants are scarce, a blue pigment, phycocyanin extracted from Spirulina, has attracted much attention as a natural blue colourant .

Microalgal pigments are considered environmentally sustainable because of the ability of microalgae to grow utilising sunlight, CO_2 and nutrients in wastewater thereby aiding in carbon sequestration and wastewater remediation. Thus, their application in textile dyeing processes may contribute to sustainable industrial development and circular bioeconomy strategies.

Types of Microbial Pigments

Microbial pigments can also be categorised based on the chemical structure, biosynthetic pathway and colour characteristics. Different classes of pigments have unique chemical and functional properties and they have an impact on their application in textiles.

Carotenoids are among the most common pigments in microorganisms, including β-carotene, lycopene, astaxanthin and canthaxanthin. These pigments usually give yellow, orange and red colours and have strong antioxidant activity due to their polyene structures (Mata-Gómez *et al*., 2014).

Melanins are dark brown or black pigments formed by oxidative polymerisation reactions. They are protective from ultraviolet radiation and oxidative stress and are appreciated by their UV-protective properties for textile applications (Gmoser *et al*., 2017).

Prodigiosin is a bacterial pigment belonging to the tripyrrole group of compounds and is a red coloured pigment produced predominantly by bacterial species such as Serratia marcescens. The pigment is known to possess antimicrobial and anticancer properties and has been explored for development of biofunctional textiles (Darshan and Manonmani, 2015).

Violacein is a violet pigment of indole origin produced by several bacteria. It displays antibacterial, antiviral and

Table 2 : Major Fungal Pigments and their Applications

Fungal Species	Pigment Produced	Colour	Textile Relevance	Reference
Monascus purpureus	Monascorubrin	Red	Natural textile dyeing	Mapari *et al.* (2010)
Aspergillus niger	Melanin	Black/Brown	UV-protective textiles	Gmoser *et al.* (2017)
Penicillium oxalicum	Anthraquinones	Yellow/Orange	Sustainable coloration	Dufossé *et al.* (2014)
Fusarium spp.	Naphthoquinones	Red/Violet	Functional dyeing applications	Velmurugan *et al.* (2010)

antioxidant activities and it is appropriate for medical and protective textiles (Venil *et al*., 2013).

Phycobiliproteins are water soluble pigments produced mainly by microalgae and cyanobacteria. These pigments are phycocyanin and phycoerythrin providing blue and red colours respectively and applications in natural dyeing and fluorescent materials (Morales-Oyervides *et al*., 2020). The following table classifies major microbial pigments based on microbial source, colour and functional properties (Table 3).

Production of Microbial Pigments

Microbial pigment production is the process of growing the pigment-producing microorganisms in controlled environment and nutritional factors for the production of pigment. In comparison to natural dyes extracted from plants, microbial pigment production has several merits: it is less time consuming, relatively simple to control, free from climatic influences and it is possible for large-scale industrial fermentation (Mapari *et al*., 2010).

Generally, microbial pigments are synthesized by submerged fermentation (SmF) or solid-state fermentation (SSF) depending on the type of microorganism and type of pigments. It is the submerged fermentation wherein the micro-organisms are grown in a liquid nutrient media containing carbon sources, nitrogen sources, minerals and growth factors. The method is widely adopted due to the critical parameters that influence the production of the pigment such as pH, temperature, aeration and agitation, which are precisely controlled in this method (Venil *et al*., 2020).

Solid-state fermentation is microbial growth on moist solid substrates with little or no free water. In solid-state systems, agricultural residues such as rice bran, wheat bran, sugarcane bagasse and fruit peels are often used as substrates. Economically, it is advantageous because the use of low-cost agro-industrial wastes, reduction of wastewater generation, and energy consumption (Pandey *et al*., 2018).

The microbial pigment production is influenced by several factors like temperature, pH, incubation period, aeration, carbon source, nitrogen source and light. The synthesis of pigments by different microorganisms necessitates specific environmental conditions for the optimum synthesis. Carotenoid production in yeasts is often increased under stress conditions such as high salinity, light exposure or nutrient limitation (Buzzini *et al*., 2017). Similarly, pigment production in bacterial systems may be increased in response to environmental stress or quorum sensing mechanisms.

Downstream processing techniques like solvent extraction, centrifugation, filtration, chromatography and membrane separation are used to recover pigments and purify them from culture broth after fermentation. The efficiency of extraction depends on the pigment being intracellular or extracellular. Cell disruption techniques like sonication, homogenisation or enzymatic lysis are typically needed before extraction with solvent to release intracellular pigments (Singh *et al*., 2021).

The development of biotechnology and metabolic engineering has significantly contributed to the microbial pigments production. The yield and the cost of production has been improved by genetic engineering of pigment producing strains, optimisation of fermentations parameters and utilization of cheap renewable substrates. The achievements have enhanced the industrial application of microbial pigments for sustainable textile dyeing and other commercial applications.

Industrial Production and Biotechnological Advances in Microbial Pigments

Because the synthesis of microbial pigments is possible with the use of controlled cultivation under a wide range of conditions, the production of microbial pigments has become highly relevant both in science and industry. The microbial pigments have several benefits over natural colours extracted from plants because the production cycles are quick, seasonal variations are not a problem, scaling up is easy, and fermentation technologies can enable continuous production (Mapari *et al*., 2010). Microbial pigment production is now much

Table 3 : Classification of Major Microbial Pigments

Pigment Type	Major Colour	Microbial Source	Functional Properties	Reference
Carotenoids	Yellow–Red	Bacteria, yeasts, algae	Antioxidant activity	Mata-Gómez *et al.* (2014)
Melanin	Brown–Black	Fungi, bacteria	UV protection	Gmoser *et al.* (2017)
Prodigiosin	Red	*Serratia* spp.	Antimicrobial activity	Darshan and Manonmani (2015)
Violacein	Violet	*Chromobacterium* spp.	Anticancer and antioxidant properties	Venil *et al.* (2013)
Phycocyanin	Blue	Cyanobacteria	Natural blue colorant	Morales-Oyervides *et al.* (2020)

more feasible for large scale production in applications for textile dyeing, due to recent developments in industrial biotechnology, microbial engineering and fermentation science.

Typically, microbial pigments are produced via biotechnological fermentation processes, in which specific microorganisms are grown in nutrient rich media under controlled environmental conditions. The synthesis of pigment molecules inside and outside the microbial cells depends on the microbial species and type of pigment molecules produced during growth and metabolism of the microbes. The conditions of fermentation such as composition of nutrients, pH, temperature, aeration, and fermentation time all have a significant effect on the production yield and quality of pigments (Venil *et al*., 2020).

Selection of appropriate microorganisms is one of the key factors of pigment production as different microbial strains produce pigments of different colours, chemical structures and functional properties. Bacteria, fungi, yeasts and microalgae have been explored as industrial pigment producers because of their metabolic diversity and their ability to synthesise bioactive compounds with commercial value (Ramesh *et al*., 2019). Also, microbial fermentation systems can use renewable substrates and agro-industrial wastes, which are advantageous for sustainable production and waste valorisation.

Microbial pigments industrial production generally involves three major stages, *i.e.*, fermentation and microbial cultivation, optimisation of the pigment biosynthesis condition and downstream processing including extraction and purification. Efficient control of these stages is critical to improve pigment yield, colour stability and economic feasibility of industrial-scale production (Fig. 1).

Fermentation Processes

Fermentation is a key factor in microbial pigment production since it provides favourable environmental and nutritional conditions for microbial growth and pigment biosynthesis. Microbial fermentation processes are generally classified as submerged fermentation (SmF) and solid-state fermentation (SSF) that have been widely studied for industrial pigment production (Pandey *et al*., 2018).

Submerged fermentation is the most common technique for the production of microbial pigments in commercial scale. In this process the microorganisms are grown in a liquid nutrient medium, containing carbon sources, nitrogen sources, minerals, vitamins and other growth-promoting substances. The fermentation broth is constantly stirred and aerated to ensure even distribution of nutrients and oxygen. Submerged fermentation has several advantages such as easy monitoring and control of the process parameters like pH, temperature, dissolved oxygen, agitation speed etc. (Venil *et al*., 2020).

Submerged fermentation is usually used for the production of bacterial pigments such as prodigiosin, violacein and carotenoids because the bacterial species

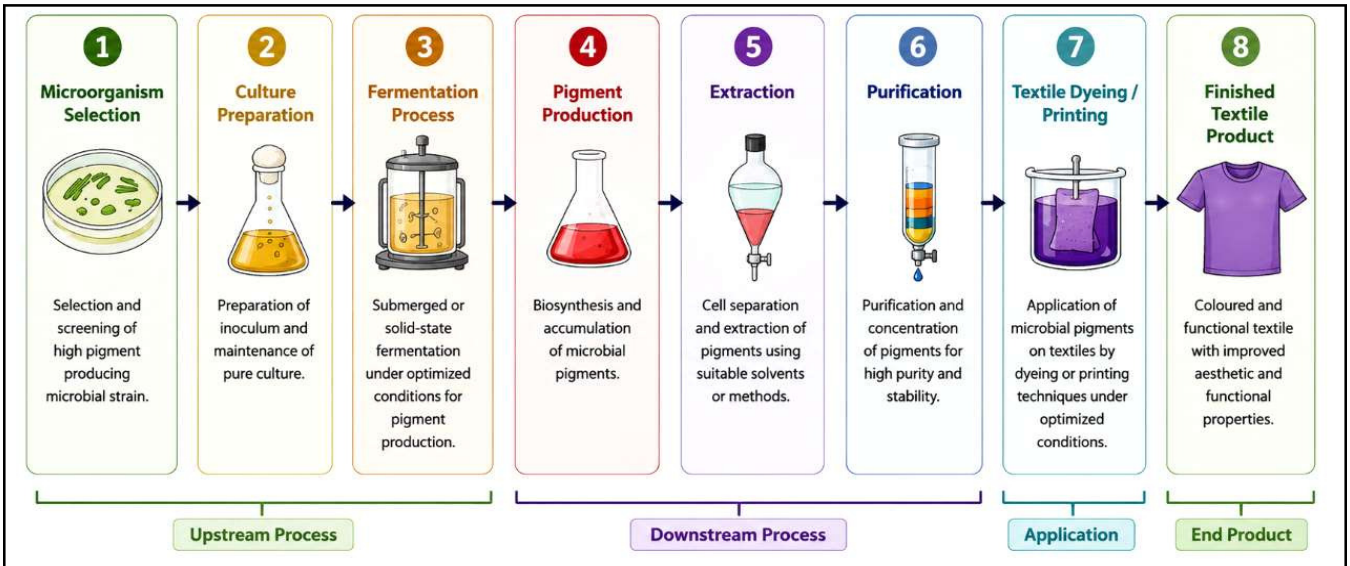

Fig. 1 : Production Process of Microbial Pigments for Textile Dyeing

generally require well-aerated liquid culture systems for efficient growth and metabolite synthesis. Similarly, yeasts and microalgae are often grown in liquid bioreactors, where the availability of nutrients and environmental conditions can be very well controlled (Buzzini *et al*., 2017).

Solid-state fermentation is the growth of microorganisms on moist solid substrates in the absence or near absence of free water. Agricultural residues such as rice bran, wheat bran, fruit peels, sugarcane bagasse and maize husk are the most commonly used substrates for solid-state systems. This method is particularly appropriate for filamentous fungi because fungal hyphae are able to grow efficiently on solid substrates and to produce high amounts of pigments (Dufossé *et al*., 2014).

Solid-state fermentation has some environmental and economic advantages with reduced water consumption, energy input and use of low-cost agricultural waste materials as substrates. In addition, solid-state systems usually result in higher pigment concentrations when compared to submerged fermentation due to reduced dilution effects (Pandey *et al*., 2018). Monitoring the process and controlling the parameters are, however, generally more difficult in solid-state systems due to possible variations in the distribution of temperature, moisture and aeration within the substrate matrix.

Recent advances in bioreactor design and fermentation technology have improved the efficiency of microbial production of pigments. Depending on the microorganism and the production requirements, stirred-tank bioreactors, airlift bioreactors, packed-bed reactors and photobioreactors are increasingly being used. Photobioreactors are especially relevant for the production of microalgal pigments, due to the controlled light exposure, which is crucial for photosynthetic growth and pigment biosynthesis (Morales-Oyervides *et al*., 2020). The following table presents major fermentation methods used for microbial pigment production and their industrial significance (Table 4).

Factors Affecting Pigment Production

Microbial pigment production is highly influenced by a spectrum of physical, chemical, nutrient and environmental factors regulating microbial metabolism and biosynthesis of secondary metabolites. Optimisation of these parameters is important for the improvement of pigment yield, productivity, colour intensity and process efficiency during industrial scale fermentation (Ramesh *et al*., 2019).

Temperature is one of the most important parameters affecting growth of microorganisms and biosynthesis of pigments. The optimal temperature for pigment production depends on the microorganism, since the enzymatic activities involved in metabolic pathways are temperature-dependent. Generally, moderate temperature is favourable for pigment synthesis while very high or low temperatures can inhibit the growth of microbes and reduce pigment accumulation (Venil *et al*., 2013). Thus, pigment formation by Serratia marcescens, *i.e*. production of prodigiosin, is often stimulated at temperatures of about 28–30°C, whereas higher temperatures may be inhibitory.

The pH of the culture media also has a significant effect on microbial pigment production due to its influence on the solubility of nutrients, enzyme activity, membrane transport and microbial metabolism. The optimum pH for biosynthesis of pigments differs with the different microorganisms. In general, a slight acidic or neutral pH is found to be favourable for fungal and yeast pigment production, while some bacterial systems require alkaline pH to produce maximum pigment (Mapari *et al*., 2010).

Sources of carbon and nitrogen is particularly important because they are nutrient and metabolic precursors needed for the growth and secondary metabolite production by microorganisms. The fermentation media are typically a carbon source like

Table 4 : Major Fermentation Methods Used for Microbial Pigment Production

Fermentation Method	Characteristics	Suitable Microorganisms	Advantages	Reference
Submerged Fermentation (SmF)	Liquid nutrient medium with aeration and agitation	Bacteria, yeasts, microalgae	Easy process control and scalability	Venil *et al.* (2020)
Solid-State Fermentation (SSF)	Growth on moist solid substrates	Filamentous fungi	Lower water and energy consumption	Pandey *et al.* (2018)
Photobioreactor Cultivation	Light-controlled cultivation system	Microalgae and cyanobacteria	Enhanced photosynthetic pigment production	Morales-Oyervides *et al.* (2020)
Stirred Tank Bioreactor	Mechanically agitated fermentation system	Bacteria and yeasts	Uniform mixing and oxygen transfer	Singh *et al.* (2021)

glucose, sucrose, glycerol, molasses and starch. Similarly, nitrogen sources such as peptone, yeast extract, ammonium salts and agricultural by-products have a significant impact on microbial protein synthesis and pigment formation (Singh *et al*., 2021).

Aeration and oxygen availability are of special importance for aerobic microorganisms since oxygen is involved in different metabolic reactions related to pigment biosynthesis. In submerged fermentation systems, for instance, microbial growth and pigment yield can be limited by poor oxygen transfer. Therefore, the agitation rate influences oxygen transfer efficiency, nutrients distribution and microbial cells dispersion in the fermentation broth (Buzzini *et al*., 2017).

Light exposure also affects the pigment production of several microorganisms, particularly photosynthetic microalgae and pigment-producing bacteria. Some pigments such as carotenoids and phycobiliproteins are produced as a result of light-induced oxidative stress for protective purposes. Light intensity and wavelength variations can therefore considerably change the pigment concentration and composition (Morales-Oyervides *et al*., 2020).

Another important parameter is incubation time as the synthesis of pigments by microbes is generally related to specific growth phases. Many pigments are secondary metabolites that are produced mainly in the stationary phase of microbial growth when nutrient limitation or environmental stress leads to the accumulation of metabolites (Venil *et al*., 2020). The following table summarizes important factors affecting microbial pigment production during fermentation processes (Table 5).

Extraction and Purification

Microbial pigments are extracted and purified from microbial biomass or fermentation broth by the microbial fermentation before they can be used for textile dyeing and other industrial applications. The efficiency of extraction and purification can have a significant impact on the pigment recovery, colour quality, stability and commercial viability (Singh *et al*., 2021).

The extraction method depends mainly on the nature of the pigment whether it's intracellular or extracellular. Extracellular pigments are excreted directly into the fermentation broth and are relatively easier to recover by filtration or centrifugation. However, the pigments are trapped inside the microbial cells and need to be extracted using cell disruption methods (Ramesh *et al*., 2019).

Cell disruption is done using mechanical, physical, chemical and enzymatic methods. Intracellular pigments are commonly extracted by mechanical disruption techniques, such as homogenisation, bead milling and ultrasonication, which disrupt microbial cells to break their cell wall. Solvents, detergents or acids might also be used for chemical treatments to release pigments depending on the type of microbial species and the properties of the pigment (Venil *et al*., 2020).

One of the most common methods of recovering pigments is by solvent extraction. Many microbial pigments can be dissolved by commonly used organic solvents like ethanol, methanol, acetone, ethyl acetate or hexane. The solvent is chosen according to the polarity, stability and use of the pigment. A reduction in toxicity coupled with improved sustainability has led to the use of eco-friendly solvents like ethanol (Mapari *et al*., 2010).

Table 5 : Factors Influencing Microbial Pigment Production

Factor	Influence on Pigment Production	Examples	Reference
Temperature	Regulates enzyme activity and metabolism	Higher temperatures may suppress pigment synthesis	Venil *et al.* (2013)
pH	Affects nutrient availability and metabolic activity	Neutral to slightly acidic pH favours many pigments	Mapari *et al.* (2010)
Carbon Source	Provides energy and biosynthetic precursors	Glucose and sucrose commonly used	Singh *et al.* (2021)
Nitrogen Source	Influences protein synthesis and metabolism	Peptone and yeast extract enhance production	Ramesh *et al.* (2019)
Aeration	Improves oxygen transfer in aerobic fermentation	Increased agitation enhances pigment yield	Buzzini *et al.* (2017)
Light Intensity	Stimulates protective pigment synthesis	Important in microalgal cultivation	Morales-Oyervides *et al.* (2020)
Incubation Time	Determines metabolite accumulation phase	Maximum pigments often produced during stationary phase	Venil *et al.* (2020)

The pigments are purified after extraction from the impurities, residual biomass and unwanted metabolites. Filtration and centrifugation are preliminary ways of clarifying pigment extracts. Highly purified pigments for industrial and biomedical uses are obtained by advanced pigment purification methods such as chromatography, membrane filtration, crystallisation and high-performance liquid chromatography (HPLC) (Singh *et al*., 2021).

Pigment stability is another important factor during downstream processing as heat, oxygen, light and pH fluctuations may cause pigment degradation. Therefore, stabilisation agents, encapsulation technologies, and controlled storage conditions are often used to preserve the quality and improve the shelf life of the pigment (Morales-Oyervides *et al*., 2020).

The development of green extraction technologies such as supercritical fluid extraction, ultrasound-assisted extraction, microwave-assisted extraction and enzyme-assisted extraction has been the focus of recent research to improve pigment recovery while reducing solvent consumption and environmental impact. These advanced techniques are increasingly seen as important for sustainable industrial production of microbial pigments for textile uses.

Application in Textile Dyeing

The use of microbial pigments for textile dyeing is attracting more and more attention with the increasing demand of environmentally sustainable colouration technologies. Conventional synthetic dyes used in textile industries are associated with hazardous chemicals, non-biodegradable compounds and generate highly polluted wastewater. However, microbial pigments are an environment friendly alternative as they are biodegradable, renewable and can be produced using sustainable biotechnological techniques (Venil *et al*., 2020). Besides colour, many microbial pigments have functional properties like antimicrobial activity, UV protection, antioxidant characters and bioactivity which can improve the performance and value of textile products.

The pigments of microorganisms have been successfully applied for dyeing various textile materials like cotton, wool, silk, polyester, nylon and blended fabrics. In textile processing their application generally includes extraction of pigments from microbial biomass and then dyeing or printing under optimised processing conditions. The dyeing performance of microbial pigments depends on various factors such as the chemistry of the pigment, structure of the fibre, affinity of the dye to the fibre, mordanting conditions, pH, temperature.

Dyeing Mechanism

The microbial pigment dyeing mechanism is based on interaction of the pigment molecules with textile fibres by different physical and chemical forces. Microbial pigments, similar to conventional dyes, must be able to penetrate the fibre structure and have sufficient affinity to the textile material in order to form a stable and durable colour. Many factors influence the interaction of dye with fibre like structure of pigment, composition of fibre, conditions of dyeing, pH, temperature and presence of mordants or binding agents (Shahid and Mohammad, 2013).

Microbial pigments can interact with textile fibres by the hydrogen bonding, van der Waals force, ionic force, hydrophobic force and covalent force depending on the type of pigment and the surface of the fibre. Natural fibres like cotton, wool and silk have –OH, –NH and –COOH groups which have a reactive character and thus can absorb and fix the pigment. However, synthetic fibers such as polyester and nylon typically have less hydrophilicity and fewer reactive groups that may decrease the efficiency of dyeing uptake and could need different dyeing conditions (Venil *et al*., 2020).

Many microbial pigments have an affinity to protein fibres due to the presence of amino acid residues in protein-based materials that can strongly interact with the pigment molecules. For example, the dyeing behaviour of wool and silk fabrics with pigmented compounds such as prodigiosin, carotenoids, Monascus pigments has been found to be quite effective with improved hydrogen bonding and hydrophobic interactions (Mapari *et al*., 2010). Cellulosic fibres such as cotton often require mordanting agents or pretreatment processes to improve the properties of pigment fixation and colour fastness.

Mordants are chemicals that help pigments to bind better to textile fibres and improve colour durability. In the traditional natural dyeing systems, metal salts such as alum, ferrous sulphate and copper sulphate have been used as mordants. Nevertheless, due to environmental issues related to metallic mordants, recent studies have focused more on bio-mordants and eco-friendly methods of fixation by tannins, enzymes and biopolymers (Yusuf *et al*., 2017) (Fig. 2).

The dyeing process usually consists of preparation

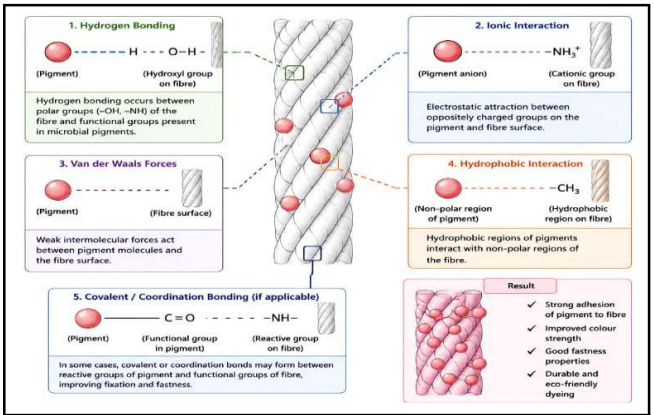

Fig. 2 : Mechanism of Microbial Pigment Dyeing on Textile Fibres

of pigment extract, adjustment of dye bath conditions, dipping of textile material, heat under control and washing after dyeing to remove unfixed pigments. The dyeing efficiency will depend on dye concentration, liquor ratio, addition of pH, dyeing temperature and dyeing time. To achieve uniform colouration and improved fastness performance. These conditions need to be optimized (Singh *et al*., 2021).

Recently, scientists have investigated new dyeing methods like ultrasonic dyeing, microwave-assisted dyeing, enzymatic pretreatment and nano-encapsulation technologies to increase the dyeability and stability of microbial pigments on textiles. These strategies can be used to enhance the penetration of pigments, minimize energy use and make the textile colouration processes more environment-friendly (Kate *et al*., 2021).

Dyeing of Natural Fibres

Natural fibres are one of the most suitable substrates for microbial pigment dyeing due to the presence of functional groups which can interact effectively with natural colourants. Dyes are absorbed and fixed on to fibres such as cotton, wool, silk, flax and jute which contain hydroxyl, amino and carboxyl groups. As a result, several studies have demonstrated the successful application of microbial pigments on natural textile materials with acceptable colour strength and functional performance (Venil *et al*., 2013).

Cotton is one of the most used natural fibres in textile industries and is studied widely in microbial pigment dyeing. The fibre is largely made up of cellulose and therefore contains hydroxyl groups that can hydrogen bond with pigment molecules. However, some microbial pigments have relatively low affinity to cellulose fibres, thus, mordanting or fibre pretreatment is required to improve dye fixation and fastness properties (Yusuf *et al*., 2017). Several studies report successful dyeing of cotton fabrics with bacterial pigments such as prodigiosin and fungal pigments from Monascus species .

Cellulosic fibres are less susceptible to microbial pigments than protein fibres such as wool and silk as amino acid residues in these fibres are more capable of forming stronger ionic and hydrogen bonding interactions with pigment molecules. Amino, carboxyl and disulphide groups are present in wool fibres which provide effective adsorption of pigments and better colour fastness. Silk has good dye uptake due to its proteinaceous nature and smooth fibre morphology (Mapari *et al*., 2010).

The multifunctional performance of various microbial pigments on natural fibres apart from the colouration property has been demonstrated. Such as cotton fabrics dyed with prodigiosin have been found to have antibacterial activity against pathogenic microorganisms, and cotton fabrics treated with melanin have proved to be ultraviolet protective (Ravikumar *et al*., 2022). This kind of multifunctional properties are especially useful for medical textiles, sportswear and protective clothing use.

The use of microbial pigments to dye natural fibres also has considerable environmental benefits. In general microbial pigment dyeing needs lower concentrations of hazardous chemicals and generates less toxic wastewater than synthetic dyeing systems. Further, the biodegradable nature of microbial pigments diminishes the environmental persistence and ecological toxicity of textile effluents (Niinimäki *et al*., 2020). The following table presents the application of microbial pigments on natural textile fibres and their associated functional properties (Table 6).

Dyeing of Synthetic Fibres

Microbial pigments on synthetic fibres have also attracted significant interest as synthetic textiles such as polyester, nylon, acrylic and polypropylene constitute a large percentage of worldwide textile production. However, the dyeing of synthetic fibres with microbial pigments is more complicated than natural fibres because of the less hydrophilic nature, more compact molecular structure and less reactive functional groups for the binding of pigments of synthetic polymers (Singh *et al*., 2021).

Polyester is one of the most prevalent synthetic fibre

Table 6 : Application of Microbial Pigments on Natural Fibres

Fibre Type	Microbial Pigment	Microbial Source	Functional Properties	Reference
Cotton	Prodigiosin	*Serratia marcescens*	Antimicrobial activity	Venil *et al.* (2013)
Wool	Monascus pigments	*Monascus purpureus*	Good colour fastness	Mapari *et al.* (2010)
Silk	Violacein	*Chromobacterium violaceum*	Antioxidant properties	Ravikumar *et al.* (2022)
Cotton	Melanin	Fungal species	UV protection	Gmoser *et al*. (2017)
Linen	Carotenoids	Yeast species	Natural coloration	Buzzini *et al.* (2017)

that is used due to its durability, resistance to wrinkle and mechanically strong. But the surface of the polyester fibres is extremely hydrophobic and natural dyes do not bind to it. It is therefore necessary to use higher dyeing temperatures, use of a carrier agent, surface modification or nano-finishing to enhance the microbial pigment uptake on polyester fabrics (Kate *et al*., 2021). Some of the carotenoids and prodigiosin pigments showed moderate affinity towards polyester fibres under optimised dyeing conditions.

The dyeability of nylon fibres is comparatively better since the polyamide structure contains amino groups which can interact with pigment molecules by ionic and hydrogen bonding. Several microbial pigments such as violacein, Monascus pigments and bacterial carotenoids have been successfully applied on nylon fabrics with acceptable colour strength and wash fastness properties (Venil *et al*., 2020).

Recent studies have focused on the improvement of the microbial pigment application on synthetic fibres by using fibre surface modification techniques such as plasma treatment, enzymatic treatment, ozone treatment and nano-coating technologies. These methods increase the roughness of the fibre surface and create reactive functional groups which improve the adsorption and fixation of pigments (Ramesh *et al*., 2019).

Encapsulation technologies have also been proposed as promising approaches to improve the stability of microbial pigments and their dyeing performance on synthetic textiles. Microencapsulation and nanoencapsulation techniques help to protect the pigments from degradation due to the heat, light and oxidation and also help in the controlled release and dye-fibre interaction (Singh *et al*., 2021).

However, there are still some issues like colour fastness and industrial scalability, but the feasibility of microbial pigment dyeing of synthetic textile materials is still enhanced by the progress of textile finishing technologies and biotechnological innovations.

Printing Applications

Apart from dyeing, microbial pigments have shown promising potentials for textile printing applications. Textile printing is the application of colour to fabric in specific areas to produce decorative designs and patterns. Conventional textile printing systems usually use synthetic pigments, binders, thickeners and solvents which can cause environmental pollution and the production of toxic wastewater. Therefore, the microbial pigments applied for textile printing are a sustainable alternative for eco-friendly textile designing and surface colouration (Dufossé *et al*., 2014).

Microbial pigments can be incorporated in printing pastes comprising natural or synthetic binders, thickeners and fixation agents, prior to printing on textile substrates by screen printing, block printing, inkjet printing or digital textile printing methods. Rheological properties and stability of the pigment formulation are important factors affecting print quality, colour uniformity and fixation efficiency (Venil *et al*., 2020).

Several studies have shown the effectiveness of bacterial and fungal pigments in textile printing systems. Pigments like prodigiosin, violacein, Monascus pigments and carotenoids have been used in the development of bright printed patterns on cotton, silk and blended fabrics (Ravikumar *et al*., 2022). Sometimes, printed textiles also revealed additional antimicrobial and antioxidant functionalities deriving from the biological activity of the pigments .

Digital textile printing technologies have further enhanced the potential for microbial pigment application as digital systems require less amount of water, less chemical usage and precise colour placement in comparison to conventional printing methods . Bio-inks prepared with microbial pigments are being increasingly studied for sustainable digital textile printing applications (Kate *et al*., 2021).

A major problem in microbial printing is the stability of pigments in the storage and fixation and washing steps. Some pigments are prone to heat, ultra-violet light and

oxidation causing fading with time. So, continuing studies are conducted on better pigment stabilization techniques like encapsulation methods, crosslinking agents, and environmental friendly binders to further improve the durability of the printing and colour fastness of the pigment (Singh *et al*., 2021).

The use of microbial pigments in textile printing is in line with the principles of sustainable textile production as it can contribute to a decrease in chemical pollution, to biodegradable colouring systems and environmentally friendly production technologies.

Functional and Sustainable Properties

Microbial pigments have a number of functional and sustainable properties to be promising alternatives to conventional synthetic dyes in textile applications. Besides natural colour, many microbial pigments possess antimicrobial, antioxidant, UV protective and bioactive properties which can improve the performance of textile materials. Such multifunctional properties have increased the scientific interest of microbial pigments for medical textiles, protective clothing, sportswear and sustainable fashion products (Venil *et al*., 2013). Furthermore, microbial pigments are generally biodegradable, produced from renewable biological resources via environment friendly fermentation processes and have a relatively lower ecological impact compared with synthetic dyes (Mapari *et al*., 2010).

Antimicrobial Properties

A number of microbial pigments have been identified with high antimicrobial activity against pathogenic microorganisms like bacteria and fungi. Some of the pigments such as prodigiosin, violacein and melanin can inhibit microbial growth by disrupting cell membranes, inducing oxidative stress and interfering with microbial metabolic pathways (Ravikumar *et al*., 2022). Therefore, textile materials dyed by these pigments can be used for providing extra hygienic and protection functions, particularly in healthcare textiles, wound dressings, sportswear, antimicrobial garments etc. Microbial pigments can also help in controlling the formation of the odours and microbial contamination during textile use.

UV Protection

Some natural microbial pigments are also found to have the ability to absorb UV, which can be used to improve the UV properties of textile materials. Pigments like melanin and carotenoids are able to absorb UV radiation, to neutralise reactive oxygen species and to protect microorganisms from damage caused by UV radiation and other oxidants (Gmoser *et al*., 2017). Applied to fabrics, these pigments can help to reduce the Ultra Violet transmission and enhance protection properties of fabric products used for outdoor and protection purposes. With this trend of consumer demand for UV-protective clothing, there has been further interest in microbial pigments as multi-functional textile colourants.

Biodegradability and Eco-friendliness

Microbial pigments are generally biodegradable and produced from renewable biological resources and so they are a sustainable alternative to the synthetic dyes from petrochemical. Unlike synthetic dyes that are resistant to biodegradation and tend to persist in the environment, microbial pigments can be degraded by the environmental microorganisms thus reducing the ecological toxicity and long-term pollution hazards (Mapari *et al*., 2010). Furthermore, microbial fermentation production is characterised by relatively lower dependence on hazardous chemicals and fossil resources. Moreover, microbial cultivation can use agricultural residues and industrial by-products as substrates, contributing to waste valorisation and circular bioeconomy approaches in sustainable textile manufacturing.

Reduced Environmental Pollution

Microbial pigments can be used in textile dyeing to significantly reduce environmental pollution of conventional dyeing systems. A large number of synthetic textile dyes produce heavily pigmented wastewater containing toxic chemicals, salts and hard-to-break-down organic molecules, which increase the biological oxygen demand (BOD) and chemical oxygen demand (COD) of aquatic environments (Yaseen and Scholz, 2019). On the other hand, microbial pigments are generally less toxic and biodegradable, and therefore they have a relatively lower impact on the environment. So the use of microbial pigments can lead to cleaner production processes, reduced wastewater pollution and more sustainable practices in textile processing. The Table 7 summarizes the major functional properties of microbial pigments relevant to sustainable textile applications.

The Table 8 provides a comparative analysis between synthetic dyes and microbial pigments based

Table 7 : Functional Properties of Major Microbial Pigments Used in Textile Applications

Microbial Pigment	Major Functional Property	Textile Application	Reference
Prodigiosin	Antimicrobial activity	Medical textiles and sportswear	Venil *et al.* (2013)
Violacein	Antibacterial and antioxidant properties	Functional apparel	Ravikumar *et al*. (2022)
Melanin	UV protection and antioxidant activity	Protective clothing	Gmoser *et al.* (2017)
Carotenoids	Antioxidant and UV absorption	Sustainable fashion textiles	Mata-Gómez *et al*. (2014)
Phycocyanin	Natural blue coloration and bioactivity	Eco-friendly textile dyeing	Morales-Oyervides *et al.* (2020)

Table 8 : Comparison Between Synthetic Dyes and Microbial Pigments

Parameter	Synthetic Dyes	Microbial Pigments	Reference
Source	Petrochemical-based	Biological and renewable	Mapari *et al.* (2010)
Biodegradability	Often non-biodegradable	Generally biodegradable	Venil *et al.* (2020)
Toxicity	May contain hazardous chemicals	Comparatively lower toxicity	Berradi *et al.* (2019)
Wastewater Pollution	High	Relatively lower	Yaseen and Scholz (2019)
Functional Properties	Mainly coloration	Colour + bioactivity	Ravikumar *et al.* (2022)
Sustainability	Limited	Environmentally sustainable	Niinimäki *et al.* (2020)

on environmental and sustainability parameters.

Challenges in Commercialization

Although microbial pigments have attracted interest as environmentally friendly substitutes to synthetic textile colourants, many technological, economic and industrial challenges still remain to their large-scale commercialisation. Microbial pigments have several major hurdles to their widespread industrial utilisation despite the significant environmental and functional benefits, including production cost, pigment stability, scalability, and regulatory approval (Venil *et al*., 2020). Commercial textile industries require the colourants to be cost effective, highly stable, reproducible and compatible to existing processing systems. Therefore, it is necessary to tackle these problems to make microbial pigments potential alternatives to traditional synthetic dyes for textile production.

Production Cost

One of the major limitations associated with microbial pigments is the relatively high production cost as compared to the conventional synthetic dyes. Industrial scale production of microbial pigments includes the cost of microbial cultivation, fermentation media, sterilisation, energy consumption, downstream processing, extraction and purification (Mapari *et al*., 2010). Special nutrient media and controlled fermentation conditions are often used for obtaining the best pigment yields. This also adds to the production cost. Moreover, extraction and purification processes often involve expensive solvents, filtration systems and chromatographic techniques which decrease economic feasibility at industrial scale.

Another contributor to the cost of microbial pigment production is the low pigment yield of some of the microbial strains. Most microorganisms naturally produce pigments in very small quantities and therefore, large-scale production is not economically feasible. Hence, there is growing interest in metabolic engineering, strain improvement, and optimisation of fermentation conditions to increase pigment productivity and decrease the production costs (Singh *et al*., 2021). The use of low cost renewable substrates and agro-industrial wastes such as molasses, fruit peels and agricultural residues has also shown potential to improve economic sustainability in microbial pigment production systems.

Stability and Fastness Issues

For commercial application of microbial pigments in textile dyeing, stability of pigment and colour fastness are important parameters. However, many natural microbial pigments are sensitive to environmental factors such as light, heat, oxygen, moisture and pH variations which can lead to colour fading or degradation during textile processing and use (Ramesh *et al*., 2019). Fastness performance is one of the important quality parameters for industrial acceptance as generally textile products are expected to have colour durability under conditions such as washing, rubbing, perspiration and exposure to sunlight.

Because the dye-fibre interactions are weaker than the synthetic dyes, and the chemical fixation is limited on the textile substrates, some microbial pigments may have relatively poor fastness properties such as wash fastness,

Table 9 : Major Stability Challenges Associated with Microbial Pigments

Challenge	Effect on Textile Application	Possible Solution	Reference
Light sensitivity	Colour fading during sunlight exposure	Encapsulation and UV stabilizers	Ramesh *et al*. (2019)
Thermal instability	Pigment degradation at high temperatures	Controlled dyeing conditions	Venil *et al.* (2020)
Poor wash fastness	Loss of colour during laundering	Mordanting and crosslinking agents	Kate *et al.* (2021)
Oxidative degradation	Reduced shelf life and colour intensity	Antioxidant stabilization systems	Singh *et al.* (2021)

light fastness and rubbing fastness in comparison to synthetic dyes. To overcome these drawbacks, the researchers are looking to the use of mordants, crosslinking agents, encapsulation technologies and fibre surface modification treatments to enhance the stability and durability of pigments (Kate *et al*., 2021). Advanced techniques such as nanoencapsulation and biopolymer assisted fixation have demonstrated good results for colour retention and for the protection of the colours against environmental degradation. The Table 9 highlights major stability and commercialization challenges associated with microbial pigments in textile dyeing applications.

Scalability Challenges

The production of microbial pigments at laboratory scale has shown promising results, however, the scale-up of these processes for industrial textile applications is still a major challenge. Industrial-scale fermentation requires process consistency, efficient oxygen transfer, large bioreactor systems, contamination prevention and optimisation of nutrient utilisation which increase process complexity (Buzzini *et al*., 2017). Differences in the growth conditions of the micro-organisms can lead to differences in the pigment yield, colour uniformity and product quality making it difficult to standardise during large scale production.

Another major scalability issue is related to downstream processing, since the extraction and purification of microbial pigments at industrial scale implies a considerable amount of energy, solvent consumption and investment in equipment. Furthermore, in textile colouration applications, where uniformity of shade and reproducibility are key quality requirements, consistency of pigments between different batches of production is also important (Venil *et al*., 2020). Thus, researchers are looking at advanced bioprocess engineering approaches, including continuous fermentation systems, immobilised cell technology and automated bioreactor monitoring, to improve scalability and industrial process efficiency.

Regulatory Concerns

In addition, regulatory approval and safety assessment are additional hurdles in the commercialisation of microbial pigments for textile applications. Many microbial pigments are environmentally friendly, but some microorganisms may produce toxic metabolites, allergens or unwanted secondary compounds which need careful assessment before industrial use (Dufossé *et al*., 2014). Thus, regulatory agencies require extensive toxicity testing, environmental risk assessment and product safety validation before microbial pigments can be approved for commercial textile processing.

Another major concern is the usage of genetically modified organisms (GMOs) in pigment production systems. Although there has been improvement in pigment yield and productivity in metabolic engineering and synthetic biology, regulatory barriers in the use of genetically engineered microorganisms may hinder its industrial implementation in some countries (Singh *et al*., 2021). Moreover, international textile industries are required to comply with environmental regulations, eco-label certifications and consumer safety standards on textile chemicals and dyes. Hence, there is a need to develop standardised production protocols, quality control systems and regulatory frameworks to support the safe and sustainable commercialisation of microbial pigments in textile industries

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Recent Advances and Future Directions

In recent years, the dyeing of textiles with microbial pigments has been enhanced by the development of biotechnology and nanotechnology, along with the use of biodegradable processing methods. Production system optimization, pigment stability, dye-fibre interaction and cost reduction of the entire process are increasingly being investigated to help large-scale commercialization. The ability of microbial pigments to be utilized as sustainable alternatives to synthetic dyes used in textile industry has been broadened with innovations in microbial strain engineering, fermentation optimization, nanoencapsulation and green processing technologies (Singh *et al*., 2021). These developments are likely to have a significant impact

on the promotion of environmentally responsible textile colouration systems and towards circular bioeconomy approaches within the textile sector.

Genetic Engineering Approaches

Due to limitations like low yield and irregular production of pigment, genetic engineering and metabolic engineering have become important approaches to enhance the microbial pigment production. By genetic modification techniques, the researcher can alter the biosynthetic pathway that produces the pigment to increase the concentration of pigment, colour intensity and production efficiency (Venil *et al*., 2020). Advances in the recombinant DNA technology and synthetic biology techniques have also allowed transferring pigment-producing genes to fast-growing microbes that are able to produce pigments under optimized industrial conditions.

Engineering bacterial, fungal and yeast strains for enhanced tolerance to environmental stress and enhanced substrate utilization efficiency in fermentation has been a focus of recent studies. Genetic modification also can decrease the production of undesirable by-products and enhance the purity and stability of the pigment (Ramesh *et al*., 2019). The use of such techniques will be expected to enhance the economic viability and industrial scale of microbial pigment production for textile use.

Nanotechnology Integration

In recent years, nanotechnology has received considerable interest in microbial pigment studies, since the property of stability, colour strength, durability and controlled release of pigments can be enhanced in textile applications by incorporating nano based system. Nanoencapsulation techniques in which the microbial pigments are encapsulated in nano carriers like liposomes, polymeric nanoparticles and nanofibres, which protects the pigments from degradation by heat, oxidation, UV radiation and environment (Kate *et al*., 2021). These can improve colour fastness and also microbial pigments' long-term stability on textile substrates. Also, nanotechnology can be used to increase surface area and to promote pigment penetration into the textile material to facilitate interactions between dye and fibre. Additionally, microbial-pigment dyed nano-finished textiles can have multifunctional properties such as antimicrobial properties, UV protection and self-cleaning properties. Hence, the use of microbial pigments and nanotechnology is a promising way to create innovative functional and sustainable textile products.

Sustainable Bioprocessing

Sustainable bioprocessing approaches are progressively investigated to reduce the environmental impact and the production cost of the microbial pigment synthesis. The researchers are interested in the use of renewable substrates, agro-industrial wastes and low-cost fermentation media to enhance economic sustainability and support waste valorisation strategies (Mapari *et al*., 2010). Great potential as nutrient sources for microbial cultivation and pigment production has been demonstrated by agricultural by-products such as fruit peels, molasses, rice bran and food processing wastes.

Bioreactor design, process automation and energy-efficient fermentation systems have been developed to further improve sustainability of microbial pigment production processes. The water consumption, energy use and waste generation during processing are investigated by using continuous fermentation systems, immobilised cell technologies and wastewater-integrated cultivation systems (Buzzini *et al*., 2017). These sustainable bioprocessing strategies are well in tune with global environmental objectives and cleaner textile manufacturing technologies.

Industrial Future Prospects

Future prospects of microbial pigments in textile industries seem to be very promising due to increasing environmental awareness and strict pollution control regulations which are in favour of adoption of sustainable colouration technologies. The increasing consumer demand for eco-friendly textiles and sustainable fashion products has further stimulated the interest in biodegradable and biologically derived textile colourants (Niinimäki *et al*., 2020). Microbial pigments have other advantages, such as multifunctionality, renewable production and lower environmental toxicity, which could increase their industrial competitiveness in the next few years.

Advanced biotechnology and textile engineering techniques are likely to be used in the future to increase pigment yield, colour consistency, fastness properties and cost-effectiveness of future industrial development. The incorporation of microbial pigments in digital textile printing, smart textiles, medical textiles and multifunctional clothes might open up new commercial possibilities for sustainable textile production (Singh *et al*., 2021). Despite

some technical and regulatory hurdles, ongoing research and technological advancements will help enable the shift to the use of bio-based microbial dyes in the future textile sector.

Summary

Microbial pigments are emerging as attractive sustainable replacements for conventional synthetic dyes because they are biodegradable, renewable and have the ability to impart multifunctional properties such as antimicrobial activity, antioxidant behaviour and ultraviolet protection. Various microorganisms including bacteria, fungi, yeasts and microalgae can be used for the production of a wide range of pigments for textile dyeing and printing processes. These pigments include carotenoids, melanin, prodigiosin, violacein and phycobiliproteins. Microbial pigments are relatively less polluting for the environment as compared to synthetic dyes and support green approaches for textile processing. Their production is based mainly on fermentation technologies followed by extraction and purification processes. pH, temperature, nutrient composition and aeration are some of the factors which significantly influence the yield and quality of the pigments.

Despite multiple environmental and functional benefits of microbial pigments, the commercial utilisation of microbial pigments is still faced with several challenges such as high production cost, low stability of pigment, limitations of fastness, scalability issues, and regulatory concerns. Recent advances in genetic engineering, nanotechnology, sustainable bioprocessing and biotechnological optimisation have shown great promise for improving pigment productivity, durability and industrial feasibility. Microbial pigments are expected to play an important role in future eco-friendly textile dyeing systems and multifunctional textile applications given the global focus on sustainable textile manufacturing and cleaner production technologies.

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